Measuring Wheat Senescence with a Digital Camera

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ABSTRACT

Documenting crop senescence rates is often difficult because of the need for frequent sampling during periods of rapid change and the subjective nature of human visual observations. The purpose of this study was to determine the feasibility of using images produced by a digital camera to measure the senescence rate of wheat and to compare the results with changes in greenness determined by two established methods. Measurements were made as part of an experiment to determine the effects of elevated CO_2 and limited soil nitrogen on spring wheat (*Triticum aestivum* L.) at the University of Arizona's Maricopa Agricultural Center, near Phoenix, AZ. "Greenness" measurements were made during senescence of the crop with a color digital camera, a hand-held radiometer, and a SPAD chlorophyll meter. The green to red (G/R) for each pixel in an image was calculated and the average G/R computed for cropped images from a digital camera representing 1 m² for each treatment and sample date. The normalized difference vegetation index (NDVI) was calculated from the red and near-infrared canopy reflectances measured with a hand held radiometer. A SPAD reading was obtained from randomly selected flag leaves. All three methods of measuring plant greenness showed similar temporal trends. The relationships between G/R with NDVI and SPAD were linear over most of the range of G/R. However, NDVI was more sensitive at low values than G/R. G/R was more sensitive above G/R values of 1.2 than SPAD because the upper limits of SPAD measurements were constrained by the amount of chlorophyll in the leaf, while G/R responded to both chlorophyll concentration in the leaves as well as the number of leaves present. Color digital imaging appears useful for quantifying the senescence of crop canopies. The cost of color digital cameras is expected to decrease and the quality and convenience of use to improve.

The rate of senescence during maturation of grain crops such as wheat can be affected by fertility levels, water stress, temperature, cultivar, and other factors (Idso et al., 1980; Seligman et al., 1983; Johnson and Kanemasu, 1983; Baret and Guyot, 1986; Frederick and Camberato, 1995). While the senescence of a crop is often conspicuous in the field, documenting the process is difficult because of the need for frequent sampling during periods of rapid change and the qualitative nature of visual observations. Non-destructive methods for determining the status of plants in the field allow more frequent data collection than plant harvesting techniques and facilitate repeated measurements on the same plants. For small plot areas, greenness can be measured non-destructively with a SPAD-502 meter from the Minolta Camera Co., Ltd, Japan, which is sensitive to photosynthetic pigments of individual leaves, or remotely with radiometers which measure reflectance of the entire plant canopy.

The SPAD-502 measures the amount of chlorophyll in the leaf, which is related to leaf greenness, by transmitting light from light emitting diodes (LED) through a leaf at wavelengths of 650 and 940 nm. It tends to normalize the index for variables such as leaf thickness and cuticle reflectance properties which are not directly related to pigment concentration. The 650-nm light corresponds to peak chlorophyll attenuation of red light. The infrared (IR) 940-nm signal is not absorbed by chlorophyll. The signal from the silicon photo diodes used to detect the transmitted light is received by a microprocessor which linearizes the signal and calculates a unitless SPAD (Soil Plant Analysis Development) value:

$$\mathrm{SPAD} = A\left[\log\left(\frac{RC_0}{RC}\right) - \log\left(\frac{IRC_0}{IRC}\right)\right] + B \qquad [1]$$

where A and B are constants, RC and IRC are currents from red and IR detectors, respectively, with the sample in place and RC_0 and IRC_0 are currents from the red and IR detectors, respectively, without a sample (Wood et al., 1993). The original equation published by Wood et al. (1993) was missing the last parentheses. The placement was determined with data from a set of spectral transmittance data from the same leaves as those measured by the SPAD meter. Spectral bands of 10 nm centered around 650 and 940 nm (Pinter, 1998, unpublished data) were used to calculate an analog to the SPAD value and compared over the range of SPAD values observed. The correct placement of the missing parentheses is as presented in Eq. [1].

SPAD meters have been used to estimate chlorophyll concentrations and infer nitrogen status of single leaves of wheat, corn (*Zea mays* L.), and other plants (Wood et al., 1993; Blackmer and Schepers, 1995). Its use for characterizing the senescence of entire canopies is somewhat limited because, in practice, leaf selection for SPAD measurement is subject to operator bias. A large number of random observations must be averaged to reduce variability and make statistical comparisons between treatments meaningful. Additionally, we have

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Abbreviations: G/R, average green to red; NDVI, normalized difference vegetation index.

some concern that repeated measurements on the same leaves over time may affect their physiological status. The sensor head of the SPAD meter is clamped directly onto the leaf to make a measurement. Because of the physical contact with the leaf, some damage is inevitable, which can result in loss of a leaf earlier than normal.

In contrast, portable hand held radiometers measure reflected light without making contact, and the same area can be measured many times without damage. Many plants are averaged in a single measurement, avoiding point sampling problems attendant to the use of SPAD meters. Multispectral reflectance data from radiometers can be used to compute a variety of vegetative indices which are well correlated with agronomic and biophysical plant parameters related to photosynthetic activity and plant productivity (Tucker, 1979; Wiegand et al., 1991; Gallo et al., 1993; Bartlett et al., 1990; Pinter et al., 1983, 1994; Jackson and Huete, 1991). One of the most useful has been the normalized difference vegetative index (NDVI) calculated as:

$$\mathrm{NDVI} = \frac{NIR - R}{NIR + R} \quad [2]$$

where NIR is the near infrared reflectance and R is the red reflectance. The specific NIR and red wavelength range used for the calculation varies in individual studies but often corresponds to the bands used in resource monitoring satellites. Hand held radiometers are available commercially with filters matching SPOT or LANDSAT systems. While NDVI theoretically can have values in the range of $-1 \leq \mathrm{NDVI} \leq 1$, NIR over almost all land surfaces is greater than R which restricts the values to the range of $0 \leq \mathrm{NDVI} \leq 1$. Idso et al. (1980) used NDVI to follow senescence rates of barley (*Hordeum vulgare* L.), durum wheat (*Triticum durum* Desf.) and spring wheat.

The development of low cost digital cameras which use CCD (charge coupled device) arrays to capture images offer an additional method for assessing greenness of crops as well as other parameters that can be sensed remotely. Dymond and Trotter (1997) used a CCD array to obtain color images of forest and pasture targets from aircraft. They were able to calibrate the camera system and use it to evaluate the bidirectional reflectance properties of different targets. Clarke (1997) used a pair of black and white CCD cameras with filters in combination with a thermal imaging system in an aircraft mounted system to estimate water stress in muskmelon (*Cucumis melo* L.). Both of these systems demonstrated that CCD array cameras can produce data that can be used to assess the status of plants in the field.

The purpose of this study was to determine the feasibility of using images produced by a digital camera priced under $1000 to measure the rate of senescence of wheat and to compare the results to changes in greenness determined using NDVI calculated from canopy measurements made with a hand held radiometer and with those using a SPAD meter on individual leaves. In this application, the rate of senescence is synonymous with the loss of greenness from the canopy which is correlated with the maturation of the crop.

MATERIAL AND METHODS

Greenness measurements were made on field plots in an experiment to determine the effects of elevated CO_2 and limited soil nitrogen on spring wheat (cv. Yecora Rojo) at the University of Arizona's Maricopa Agricultural Center, approximately 40 km south of Phoenix. A Free-Air CO_2 Enrichment (FACE) facility (Hendrey, 1993) was used to expose a wheat crop to enriched (FACE, 200 μmol mol^{-1} above ambient) and ambient (Control, ~370 μmol mol^{-1}) CO_2 levels. The experiment was replicated four times. Each replicate contained a FACE and control array equipped with blowers. Replicates were divided into a high nitrogen (350 kg ha^{-1} of N) and low-N (15 kg ha^{-1} of N) half. Fertilizer (NH_4NO_3) was applied to the plots through drip irrigation tubing positioned ~0.2 m below the soil surface, spaced 0.5 m apart and emitter holes were space 0.3 m apart. The tubing of the irrigation system extended across whole replicates such that the high-N sides of both the FACE and Blower plots shared the same tubes, and likewise for the low-N sides. Therefore, the experimental design was a strip-split plot, such as that used previously with differential irrigation treatments (Pinter et al., 1996).

Plots were irrigated after 30% of the available water in the rooted zone was depleted with an amount to replace 100% of the potential evapotranspiration since the last irrigation, adjusted for rainfall (Fox et al., 1992). The cumulative season irrigations were 623 and 550 mm in the high- and low-N treatments, respectively. They would have been nearly identical except that the last irrigations on the low-N treatment were curtailed because of the earlier maturity of the N-stressed plants. The seasonal rainfall was 22 mm before the cutoff of CO_2 and an additional 13 mm was received between the cut off of CO_2 and final harvest.

The crop was planted on 15 Dec. 1996 in east-west oriented rows spaced ~0.25 m apart and harvested on 28 May 1997. The average date of heading in the low-N plots was 22 Mar 1997 and 25 Mar 1997 in high-N plots. FACE plots were enriched 24 h per day with CO_2 from emergence (3 Jan 1997) until physiological maturity (15 May 1997). Images were taken periodically from 17 Apr 1997 until 19 May 1997 with a color digital camera (Model D-300L, Olympus America Inc., Melville, NY). The camera was positioned 1.6 m above the top of the plant canopy in the same spot in an undisturbed final harvest area of each plot in Reps 1 and 4. Images were also taken of an Ambient site in an undisturbed area midway between the blower and FACE treatment arrays in Reps 1 and 4 in both the high- and low-N strips. The camera, which had a 1:2.8 5 mm lens, always had a nadir view of the crop. The nominal field of view of the camera was 57 by 42°. Images were acquired between 1100 and 1130 h MST (solar zenith angles ranging from of 31–23°). The camera's built in supplemental flash was used for each image and a white plate with red, green, and blue strips was included at the edge of each scene to provide color balance and brightness control.

The camera produced images of 1024 by 768 pixels. Successive quadruplicates in the CCD array recorded 8-bit bands of yellow, magenta, cyan, and green visible light. There was a self focusing mechanism built into the camera. The data from the four color bands were then used via a proprietary algorithm to construct red (R), green (G) and blue (B) digital values for each pixel in the final image which was stored in the camera in JPEG (joint photographic experts group) file format. Images were down loaded from the camera to a IBM compatible personal computer (PC) as JPEG files.

The images were converted to GIF (graphic interchange file) format with Paint Shop Pro version 3.12 (JASC Inc., Minnetonka, MN). This conversion resulted in a reduction of the number of colors in the images to a maximum of 256

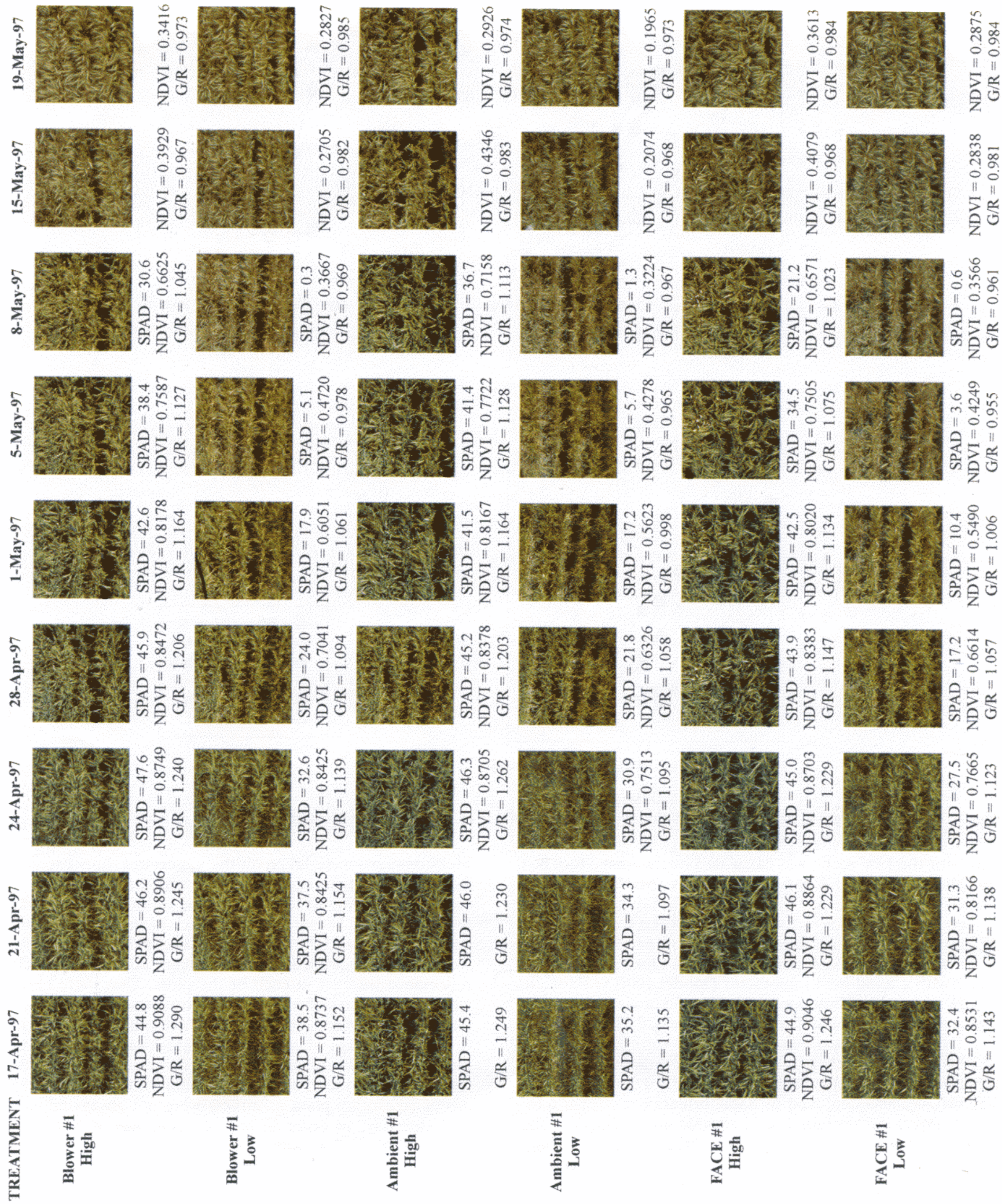

Fig. 1. Changes in greenness of wheat grown under ambient and elevated CO_2 as measured with a digital camera, normalized vegetation index (NDVI), Minolta SPAD meter, and ratio of green to red in the image (G/R).

colors. The image was then cropped to 670 by 670 pixels. Cropping to this size removed the most distorted area of the image and left an image that represented 1 m^2, with each pixel representing a square 1.49 mm on a side (approximately 10% of the width of a wheat flag leaf). The field of view of the cropped image was 36°, which means the outside pixels of the processed image were 18° off nadir. The G/R value for each pixel was then calculated from the GIF image and an average

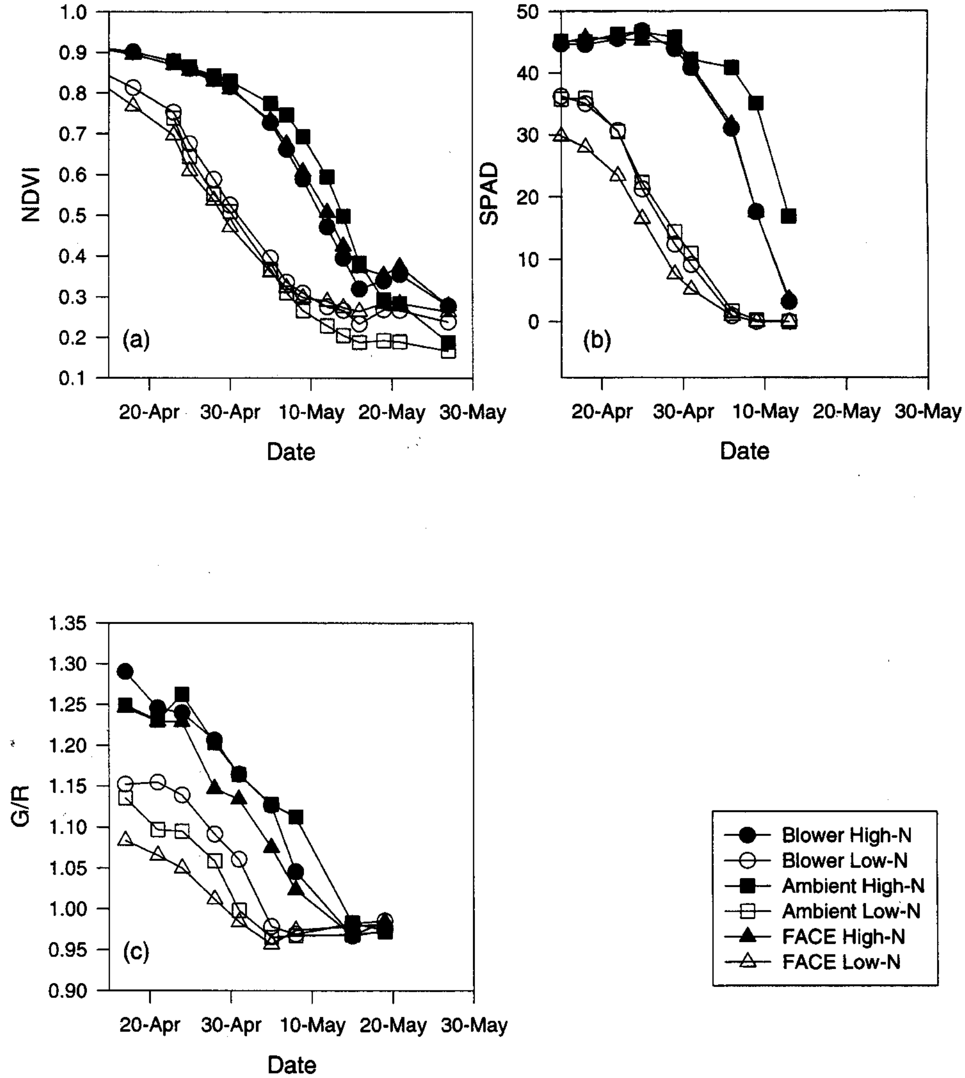

Fig. 2. Temporal changes in normalized vegetation index (NDVI), SPAD-502 values and green to red ratio (G/R) of wheat.

value for the 1 m^2 scene was calculated by the image tool box for MatLab version 4.2c.1 for Windows (MathWorks Inc., Natick, MA).

Canopy reflectance factors were measured from 6 Jan. to 27 May 1997 in the FACE and Blower plots and from 23 Apr. to 23 May 1997 in the Ambient locations. Observations were made with a hand-held radiometer (Model 100BX; Exotech, Gaithersburg, MD) equipped with 15° field of view optics and spectral filters spanning three visible (green, 0.536–0.561 μm; red, 0.61–0.68 μm; red edge 0.694–0.706 μm) and one NIR (0.79–0.89 μm) wavelength intervals. The radiometer was extended at arms length, held approximately 1.5 m above the soil surface, and deployed in nadir orientation over a 6-m-long transect that included the 1 m wide area imaged by the digital camera. Observations were centered on a morning time period corresponding to a solar zenith angle of 57° to minimize the effects of changing illumination angles on directional canopy reflectance as the season progressed. Reflectances in each waveband were calculated as ratios of reflected to incident light, as determined by frequent measurement of reflected radiation from a calibrated barium sulfate reflectance standard (Pinter et al., 1994). NDVI values were then calculated using the calibrated values of reflectance.

SPAD chlorophyll meter readings were made twice weekly in the FACE and Blower arrays from 23 Jan. to 13 May 1997 and in Ambient locations beginning on 18 Apr. 1997. Measurements were taken on the uppermost, fully expanded leaf of 18 plants in each treatment and then averaged. The meter's measuring head was clamped on the middle third of the leaf and the sensor window, which measured 2 by 3 mm, positioned to avoid the mid-rib vein where leaf widths permitted. The plants were located along the edge of the NDVI reflectance transect in each of the plots. They were chosen without regard to leaf color and no effort was made to measure the exact same plants on each date. Zero values were assigned to SPAD readings when the selected leaf was dry and brittle. This was because earlier studies had revealed anomalous performance of the meter under those conditions. The three methods of measuring plant greenness were not always taken on the same day. In order to provide estimates of NDVI and SPAD for regressions with G/R values from the digital camera, linear interpolations between dates were made. Regressions were done by the built in statistical capabilities of Sigma Plot for Windows version 2.00.

RESULTS AND DISCUSSION

As the wheat crop approached maturity, the older (lower) leaves began to senesce first, losing chlorophyll and transferring carbohydrates and protein to developing kernels in the head. Visually this process could

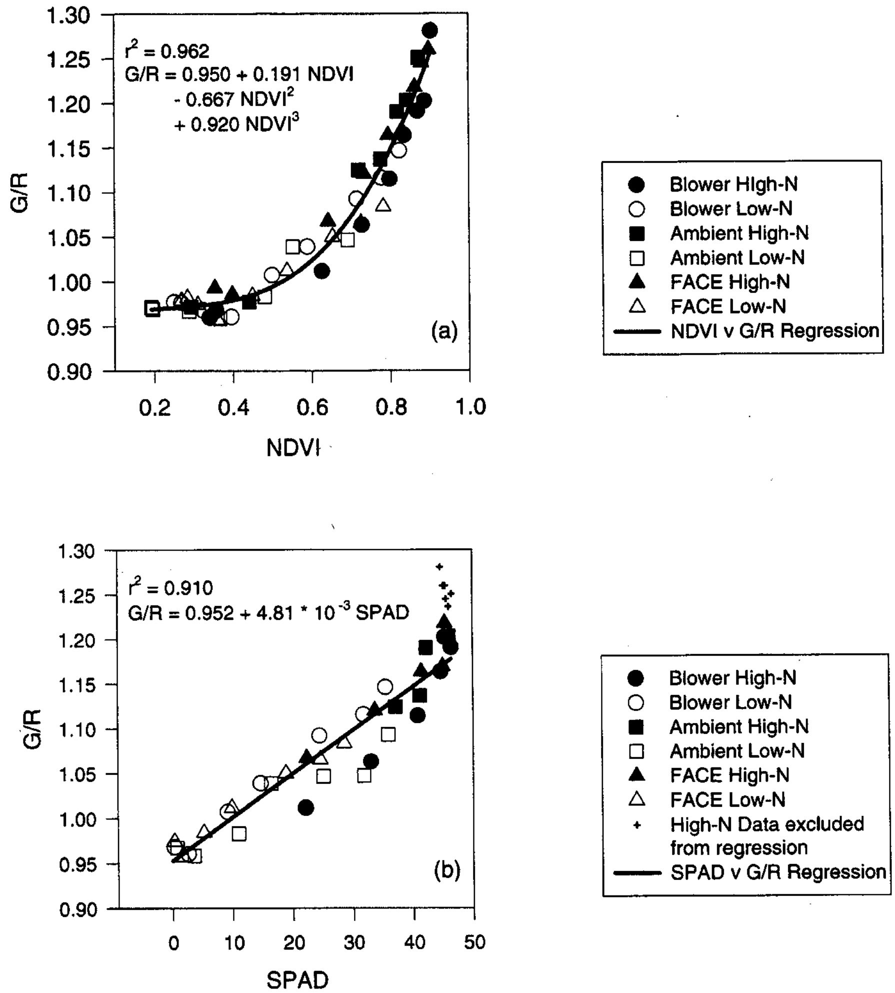

Fig. 3. The relationship of normalized vegetation index (NDVI) and SPAD-502 value to green to red ratio (G/R) for wheat.

be observed as a gradual change in canopy color from a dark green to a light yellow-brown condition. This progression of senescence was evident in the digital camera images (Fig. 1). The rate at which canopy senescence occurred was related to plant nutrient status, with plants in low-N treatments senescing about 7 to 10 d earlier than plants in the high-N treatments. The effect of CO_2 on senescence was minimal. The three methods we used for determining plant greenness (NDVI, SPAD, and G/R) had similar temporal trends and provided comparable relative rankings for the experimental treatments (Fig. 2).

Graphical representations of the data (Fig. 2) showed that each of the indices was capable of quantifying canopy senescence and detecting changes on a near-daily time step, although day-to-day variation was higher for the G/R index. The NDVI declined from April with values greater than 0.9 (typical for vigorous wheat canopies with dark green foliage, head, and awn), to less than 0.3 for a light yellow-brown canopy that was ready for harvest in late May (Fig. 2a). SPAD values ranged from 47 units for leaves in high-N treatments to 0 when leaves were senescent (Fig. 2b). The G/R index revealed that about 10 to 30% more green light than red light was being reflected by the plants at the start of our measurements (Fig. 2c). By season's end, the ratio had inverted, with slightly more red light being reflected from the canopy. Plants growing in the low-N treatments during April and early May had lower values of NDVI, SPAD, and G/R than those from the high-N treatments. For each of the indices, however, values for high- and low-N treatments tended to converge at the end of the season.

We observed differences between the indices with respect to their apparent sensitivity to experimental treatments. The G/R index for example, seemed more sensitive to high-N than low-N treatments during April. This index also showed slightly larger differences between the CO_2 treatments, FACE and Blower, than either NDVI or SPAD. It was also evident that canopy level measurements, NDVI and G/R, were more sensitive to incipient senescence of the high-N treatment than single point SPAD observations made on leaves at the top of the canopy. Some of these phenomena may be explained by considering the unique wavebands that are used in each of the greenness indices and the optical properties of the various plant tissues involved. Differences in sensor field-of-view may also have been partly responsible. The 1- by 1-m digital camera image used in the analysis, subtends an angular field-of-view of approximately 36° and thus includes proportionately more heads and awns than either NDVI (15° field-of-view) or SPAD (single point) indices.

With both wavelength and viewing geometry differences in mind, we made direct comparisons between G/R and the other two indices and found that they were comparable over much of their ranges. Overall, the relationship between G/R and NDVI was adequately described by a third order polynomial with a highly significant coefficient of determination (Fig 3a; $r^2 = 0.96$; $P < 0.001$). The relationship appeared to be nearly linear when G/R > 1. However, as the canopy approached maturity (G/R < 1), NDVI remained more sensitive to the continued decline in senescence than did the G/R index. This suggests that the functional utility of the camera imagery based on just the reflectance of light in visible wavebands may be limited late in the season. A camera system that includes NIR portions of the spectrum might show improved performance.

When G/R and SPAD data were compared, we found that the G/R index was more sensitive to the onset of senescence than SPAD. The G/R index was an integrated measure of the whole canopy greenness (multiple leaf layers, heads, and awns), while SPAD values obtained just from the green flag leaves at the top of canopy were biased estimators of the canopy. When all the data pairs were included in the regression of SPAD verses G/R, the coefficient of determination was 0.875 ($P < 0.001$) but increased to 0.910 ($P < 0.001$) when data pairs having G/R > 1.2 were excluded from the regression (Fig. 3b).

The G/R index that we developed with data from a moderate cost, uncalibrated, digital camera system provided an economical method for quantifying changes in canopy senescence that were related to our experimental treatments. Similar techniques have utility for monitoring plant response to other microclimatic factors that affect phenology, and also to genotypic differences in grain filling duration. With careful calibration, other potential uses for digital cameras include pest and pathogen detection, monitoring nutritional status, measuring green leaf area index, and estimating late-season absorption of incident photosynthetically active radiation. The cost for our digital camera system was comparable to the SPAD meter, and about 10 to 20% of that required for ground-based, calibrated radiometric approaches. Prices for CCD cameras with improved spatial resolution and on camera storage–data transfer functions continue to decrease as the digital photographic market expands. Technological advances that extend the spectral capabilities of digital cameras into the NIR portion of the spectrum and use sophisticated methods for reducing pixel-to-pixel noise are presently available on high-end systems. The data presented herein, indicate that digital cameras potentially can be used to monitor the status of crops in a timely manner and could potentially be used as a valuable management tool as more data and data analysis tools become available.

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